An outbreak of nontoxigenic corynebacterium diphtheria infection: single bacterial clone causing invasive infection among Swiss drug users

Gubler, Jacques ; Huber-Schneider, Christina ; Gruner, Eva ; Altwegg, Martin

Abstract: From 1990 to 1996, a total of 65 patients from whom Corynebacterium diphtheriae had been isolated were reported to the Swiss Federal Office of Public Health. A retrospective review of medical and microbiological records as well as results of ribotyping of available isolates was performed. Twenty-seven patients had acquired their infection without evidence of use of illicit drugs, mostly as a skin infection imported from subtropical areas (20 patients); 38 isolations were associated with intravenous drug use (IVDU) (skin, 15; respiratory tract, 10; blood, 13). Endocarditis was documented in nine patients with bloodstream infection, four of whom died. There were two additional deaths due to overwhelming sepsis. The same ribotype of nontoxigenic C. diphtheriae was found in 31 of the 32 examined isolates associated with IVDU. All non-IVDU isolates had different ribotypes. Among Swiss drug users, a single clone of nontoxigenic C. diphtheriae was found over a period of several years with a high potential to cause severe invasive infection

DOI: https://doi.org/10.1086/514997

An Outbreak of Nontoxigenic *Corynebacterium diphtheriae* Infection: Single Bacterial Clone Causing Invasive Infection Among Swiss Drug Users

Jacques Gubler, Christina Huber-Schneider, Eva Gruner, and Martin Altwegg

From the Department of Medicine, Stadtspital Triemli, and the Department of Medical Microbiology, University of Zürich, Zürich, Switzerland

From 1990 to 1996, a total of 65 patients from whom *Corynebacterium diphtheriae* had been isolated were reported to the Swiss Federal Office of Public Health. A retrospective review of medical and microbiological records as well as results of ribotyping of available isolates was performed. Twenty-seven patients had acquired their infection without evidence of use of illicit drugs, mostly as a skin infection imported from subtropical areas (20 patients); 38 isolations were associated with intravenous drug use (IVDU) (skin, 15; respiratory tract, 10; blood, 13). Endocarditis was documented in nine patients with bloodstream infection, four of whom died. There were two additional deaths due to overwhelming sepsis. The same ribotype of nontoxigenic *C. diphtheriae* was found in 31 of the 32 examined isolates associated with IVDU. All non-IVDU isolates had different ribotypes. Among Swiss drug users, a single clone of nontoxigenic *C. diphtheriae* was found over a period of several years with a high potential to cause severe invasive infection.

Corynebacterium diphtheriae is the etiologic agent of toxin-induced classic diphtheria, with involvement of respiratory membranes, the heart, and the CNS [1]. Resurgence of this disease has lately occurred in countries of the former Soviet Union [2, 3]. In the western world, outbreaks of diphtheria have been described among urban alcoholic adults [4], with a significant proportion of cases of cutaneous diphtheria.

Invasive disease due to *C. diphtheriae,* first described by Howard in 1893 [5], was a rare event until the past decade. In the past several years there has been a resurgence of reports on invasive disease, due mainly to nontoxigenic *C. diphtheriae* [6–9]. Carriage of nontoxigenic *C. diphtheriae* has been noted in special subgroups of populations, such as homosexuals in Great Britain [10], Aboriginals in Australia [11], the homeless in France [9], and drug users in Switzerland [8, 12–14]. Ribotyping has been shown to be a valuable epidemiological tool in comparison with other methods in assessing outbreaks of diphtheria in Russia [3, 15].

We report on a cluster of infections due to a single clone of nontoxigenic *C. diphtheriae* among Swiss drug users, along with a high incidence of invasive disease, occurring during the years when a permissive policy allowed an open drug scene in Zürich (''needle park'' [16]).

Methods

Patients

Diphtheria is a mandatory-reportable disease in Switzerland, although most laboratories do not routinely screen for *C. diphtheriae.* From the registry of the Federal Office of Public Health, we selected all the patients from whom *C. diphtheriae* had been isolated between 1990 and 1996. The reporting physicians were contacted and the probable source of infection was determined. For patients whose isolates were from the blood, hospital records were reviewed retrospectively. Five patients with endocarditis have been described earlier [8, 12–14]. Ten isolates were collected in a prospective study of iv drug users in Zürich [17].

Microbiology

Available isolates were identified by standard methods [18] and collected in a reference laboratory. Toxin production was determined by PCR [19]. The method of ribotyping has been described previously [13]. In short, genomic DNA was isolated, digested with two restriction nucleases (*Eco*RI and *Pvu*II; Boehringer Mannheim, Mannheim, Germany), and separated by agarose-gel electrophoresis. After Southern blotting, hybridization was done with biotin-labelled plasmid pKK3535, a pBR322-derived plasmid containing a ribosomal RNA operon of *Escherichia coli.* Hybrids were visualized with use of the BluGene Kit (GIBCO–Bethesda Research Laboratories, Gaithersburg, MD).

Results

C. diphtheriae Isolation

From 1990 to 1996, a total of 65 patients from whom *C. diphtheriae* was isolated were reported; 38 patients used iv drugs or had contact with iv drug users. Of the remaining 27 patients, 20 acquired their infection (occurring always on the skin) during travel abroad, mainly to southeastern Asia and Africa. For seven patients, history of neither travel abroad nor

Received 3 March 1998; revised 21 July 1998.

Reprints or correspondence: Dr. Jacques G. H. Gubler, Department of Medicine, Stadtspital Triemli, Birmensdorferstrasse 497, CH-8063 Zürich, Switzerland.

Clinical Infectious Diseases 1998;27:1295–8

1058–4838/98/2705–0029$03.00

Table 1. Reported isolation of *Corynebacterium diphtheriae* in Switzerland from 1990 to 1996.

Risk factor	Site of isolation	No. of isolates recovered							Total
		1990	1991	1992	1993	1994	1995	1996	
IVDU	Skin	1	4	1	1	2	4	2	15
	Nasopharynx		6	1	1				8
	Sputum	1		1					2
	Blood	2	2		3	5	1		13
Travel	Skin	2	1	2	5	3	2	5	20
CH, no IVDU	Skin		1	1					2
	Nasopharynx		1		1	1	1		4
	Blood						1		1
Total		6	15	6	11	11	9	7	65

NOTE. CH = infection acquired in Switzerland; IVDU = contact with iv drug users or use of iv drugs; travel = travel abroad to tropical and subtropical areas.

iv drug use (IVDU) could be established. The source of the isolates and risk factors are summarized in table 1.

Clinical Features of Bloodstream Infection

For 14 patients, *C. diphtheriae* was isolated from the blood. Twelve of the patients injected illicit drugs intravenously, and 10 of them frequented the open drug scene in Zürich (''needle park''). An additional patient had undergone aortic-valve replacement during childhood; although he did not use drugs himself, his girlfriend frequented the drug scene in Zürich. The last patient with bacteremia was an 83-year-old woman with bronchitis, found to have *C. diphtheriae* var. *belfanti* in her sputum and blood.

Nine patients with bacteremia had endocarditis, proven at surgery, at autopsy, or by demonstration of vegetations on echocardiography. A total of 6 patients died: 1 after emergency valve replacement, 3 of uncontrollable infection, and 2 of sepsis without evidence of endocarditis (both with concomittant *Staphylococcus aureus* bacteremia). Twelve of the 13 patients with a history of iv drug use or contact with iv drug users presented with acute septicemia. Clinical features and outcomes are shown in table 2.

Ribotyping

From the 65 patients, 38 isolates were available for ribotyping. Six isolates came from patients without a history of IVDU, and 32 isolates were from patients with a history of IVDU (including all 13 blood isolates of this group). An additional 21 strains (reference strains from collections) were included, resulting in a total of 59 strains being ribotyped.

With the exception of two isolates, all 27 reference and non-drug-related strains showed a distinct, unique restriction pattern. Among the 32 IVDU-related cases, 31 strains belonged to a single clone of nontoxigenic *C. diphtheriae* biovar. *mitis* and showed in vitro resistance to tetracycline. The remaining isolate came from a patient with a history of IVDU and clinical features of pharyngeal diphtheria in the setting of acute myeloid leukemia. Results of ribotyping are shown in table 3.

Discussion

During a limited period, a cluster of infections with a single clone of nontoxigenic *C. diphtheriae* var. *mitis* occurred in Switzerland. Although the patients who acquired their infections (in the majority of cases, on the skin) abroad harbored different strains, all but one patient with known direct or indirect contact with iv drugs were infected with one single clone of *C. diphtheriae,* as shown by ribotyping. Although the organism represented colonization of the skin or respiratory tract in most patients, a significant proportion of patients developed invasive disease. Most of the iv drug users had frequented the infamous drug scene (''needle park'' [16]) in Zürich prior to their illness. This drug scene, where easy access to drugs led to a high concentration of drug users from all of Switzerland and abroad, was dissolved in February 1995. A marked reduction, although not disappearance, of isolations of drug-related *C. diphtheriae* was noted in the following 2 years.

C. diphtheriae colonization has previously been described with regard to specific populations, e.g., urban alcoholics [4], homosexuals [20], and an aboriginal community in Australia [11]. After the occurrence of the first three cases of *C. diphtheriae* endocarditis among Swiss drug users [12, 13], Gruner and co-workers undertook a study to determine the carriage of *C. diphtheriae* among drug users in Zürich [17]. During a 10-month period in 1991–1992, they prospectively collected samples from 117 iv drug users cared for at an infirmary in Zürich. Five (4.3%) of 117 pharyngeal swabs and 5 (17.9%) of 28 swabs from superficial wounds were positive for *C. diphtheriae.* All the isolates showed the same restriction pattern by ribotyping and were included in our present study. The authors thus established that among the participants of the drug scene in Zürich, the carriage rate of this single nontoxi-

Table 2. Clinical data for 13 patients with iv drug contact and bloodstream infection with *Corynebacterium diphtheriae*.

Patient no./ age (y)/ sex	Year of presentation	Evidence of endocarditis	Involved heart valve	Preexisting heart condition	HIV status	Concomitant disorder(s)	Outcome
1/24/M	1990	E/Op/Path	Mi	None	Negative	None	Died
2/29/M	1991	E/Path	Ao	Bicuspid Ao valve status post endocarditis	Positive	None	Died
3/28/M	1991	E	Mi ?	Mild Mi insufficiency	Negative	None	Survived
4/19/F	1991	E	Mi	None	Negative	None	Survived
5/29/M	1993	E/Op	Ao	Status post 2 Ao valve replacements, prosthetic valve	Negative	Severe ARDS, DIC, renal insufficiency	Survived
6/32/F	1993	E	Ao	None	Positive	Sepsis (*Staphylococcus aureus*)	Died
7/45/M	1994	E/Path	Ao	Status post endocarditis, prosthetic Ao valve	Negative	None	Died
8/32/M	1994	No autopsy		None	Negative	Sepsis (*S. aureus*)	Died
9/25/M	1994	E	Tr	None	Negative	None	Survived
10/21/M	1994	None	None	Negative	Not tested	Aspiration pneumonia	Survived
11/27/M	1994	Path	Mi ?	None	Negative	Pneumonia, sepsis (*S. aureus*)	Died
12/32/M	1994	E	Mi ?	None	Positive	HIV infection, category B3	Survived
13/31/M	1995	E	None	Mi valve insufficiency	Unknown		Survived

NOTE. Ao = aortic; ARDS = adult respiratory distress syndrome; DIC = disseminated intravascular coagulopathy; E = echocardiographic; Mi = mitral; Op = operative; Path = pathological; Tr = tricuspid; ? = possible.

genic strain was significant and that iv drug users were available to serve as a reservoir for more invasive infections.

The propensity of the nontoxigenic strain of *C. diphtheriae* to cause invasive infection is remarkable. A review of the literature shows that until lately, invasive disease due to *C. diphtheriae* was a rare event. From the first report by Howard in 1893 [5] until 1994, only 54 cases of *C. diphtheriae* endocarditis had been reported [8]. Of the 37 cases described since 1950, only four were due to a toxigenic strain, whereas about half of the cases in the preantibiotic era had been caused by toxin-producing *C. diphtheriae.* A rise in nontoxigenic strains of *C. diphtheriae* has recently been observed, notably in Europe and Australia [9–11].

Wilson et al. [20] and Efstratiou et al. [21] reported on a marked rise in the number of strains of *C. diphtheriae* var. *gravis* sent to a reference laboratory in Great Britain, a fivefold increase in 2 years. From France, Patey and co-workers [9] recently reported the analysis of 59 isolates of *C. diphtheriae*

Table 3. Results of ribotyping of *Corynebacterium diphtheriae* isolates.

Mode of acquisition of infection	Site of isolation	No. of isolates examined	No. with ''epidemic'' ribotype	No. with other ribotypes
IVDU	Skin	12	12	0
	Nasopharynx	6	5	1
	Sputum	1	1	0
	Blood	13	13	0
Travel	Skin	4	0	4
CH, no IVDU	Nasopharynx	2	0	2
Reference strains (culture collections)	Undetermined	21	0	21
Total		59	31	28

NOTE. CH = infection acquired in Switzerland; IVDU = contact with iv drug users or use of iv drugs; travel = travel outside Europe.

causing invasive disease. The strains were collected from 1987 to 1993. In half the bacteremic patients, endocarditis was documented. Twenty-seven patients were infected by an isolate of *C. diphtheriae* var. *mitis* belonging to the same ribotype. Carriage of the organism on the skin was implicated as a major epidemiological factor, with low socioeconomic status, homelessness, and alcoholism as additional risk factors.

Populations of iv drug users as well as economically and socially deprived persons can serve as a reservoir for *C. diphtheriae.* In our series, only one case of indirect infection was documented. Given the potential of nontoxigenic strains to cause invasive infection and to acquire the toxin expressing lysogenic phage [22], this reservoir could become a serious public health problem in the future.

Acknowledgments

The authors gratefully acknowledge the help of P.-A. Raeber and the staff at the Swiss Federal Office of Public Health, as well as the reporting physicians and laboratories and the invaluable technical assistance of J. Lüthy. They thank A. von Graevenitz for careful reading of the manuscript.

References

1. MacGregor RB. *Corynebacterium diphtheriae.* In: Mandell GM, Bennett JF, Dolin R, eds. Principles and practice of infectious diseases. 4th ed. New York: Churchill Livingstone, **1994**:1865–72.
2. Hardy IRB, Dittmann S, Sutter RW. Current situation and control strategies for resurgence of diphtheria in newly independent states of the former Soviet Union. Lancet **1996**;347:1739–44.
3. Popovic T, Kombarova SY, Reeves MW, et al. Molecular epidemiology of diphtheria in Russia, 1985–94. J Infect Dis **1996**;174:1064–72.
4. Harnisch JP, Tronca E, Nolan CM, Turck M, Holmes KK. Diphtheria among alcoholic urban adults. Ann Intern Med **1989**;11:71–82.
5. Howard WT. Acute ulcerative endocarditis due to the Bacillus diphtheriae. Johns Hopkins Hosp Bull **1893**;4:32–4.
6. Lortholary O, Buu-Hoi A, Gutmann L, Acar J. *Corynebacterium diphtheriae* endocarditis in France. Clin Infect Dis **1993**;17:1072–4.
7. Tiley S, Kociuba KR, Heron LG, Munro R. Infective endocarditis due to nontoxigenic *Corynebacterium diphtheriae:* report of seven cases and review. Clin Infect Dis **1993**;16:271–5.
8. Huber-Schneider Ch, Gubler J, Knoblauch M. Endokarditis durch *Corynebacterium diphtheriae* bei Kontakt mit intravenösen Drogen: Bericht über 5 Fälle. Schweiz Med Wochenschr **1994**;124:2173–80.
9. Patey O, Bimet F, Riegel P, et al. Clinical and molecular study of *Corynebacterium diphtheriae* systemic infections in France. J Clin Microbiol **1997**;35:441–5.
10. Wilson APR. The return of *Corynebacterium diphtheriae:* the rise of nontoxigenic strains. J Hosp Infect **1995**;30(suppl):306–12.
11. Hogg GG, Strachan JE, Huayi L, Beaton SA, Robinson PM, Taylor K. Nontoxigenic *Corynebacterium diphtheriae* biovar *gravis:* evidence for an invasive clone in a south-eastern Australian community. Med J Aust **1996**;164:72–5.
12. Zuber P, Gruner E, Altwegg M, von Graevenitz A. Invasive infection with nontoxigenic *Corynebacterium diphtheriae* among drug users [letter]. Lancet **1992**;339:1359.
13. Gruner E, Zuber PLF, Martinetti Lucchini G, von Graevenitz A, Altwegg M. A cluster of non-toxigenic *Corynebacterium diphtheriae* infections among Swiss intravenous drug abusers. Med Microbiol Lett **1992**;1:60–70.
14. Anonymous. Diphtherie: Spezielle Manifestationen. Bulletin of the Swiss Federal Office for Health **1995**;42:4–5.
15. de Zoysa A, Efstratiou A, George RC, et al. Molecular epidemiology of *Corynebacterium diphtheriae* from northwestern Russia and surrounding countries studied by using ribotyping and pulsed-field gel electrophoresis. J Clin Microbiol **1995**;33:1080–3.
16. Treaster JB. Zürich: a marketplace for drugs, a bazaar of the bizarre. New York Times **1990**;Sept 27:4.
17. Gruner E, Opravil M, Altwegg M, von Graevenitz A. Nontoxigenic *Corynebacterium diphtheriae* isolated from intravenous drug users. Clin Infect Dis **1994**;18:94–6.
18. Krech T, Hollis DG. *Corynebacterium* and related organisms. In: Balows A, Hausler WJ, Herrmann KL, Isenberg HD, Shadomy HJ, eds. Manual of clinical microbiology. 5th ed. Washington, DC: American Society for Microbiology, **1991**:277–86.
19. Martinetti Lucchini M, Gruner E, Altwegg M. Rapid detection of diphtheria toxin by the polymerase chain reaction. Med Microbiol Lett **1992**;1:276–83.
20. Wilson APR, Ridgway GL, Grueneberg RN, Efstratiou A, Coolman G, Cooksen B. Routine screening for *Corynebacterium diphtheriae.* Lancet **1990**;336:1199.
21. Efstratiou A, George RC, Begg NT. Non-toxigenic *Corynebacterium diphtheriae* var *gravis* in England [letter]. Lancet **1993**;341:1592–3.
22. Pappenheimer AM, Murphy JR. Studies on the molecular epidemiology of diphtheria. Lancet **1983**;2:923–6.